U. S. DEPARTMENT OF AGRICULTURE.
BUREAU OF PLANT INDUSTRY—BULLETIN NO. 44.
B. T. GALLOWAY, *Chief of Bureau.*

THE BITTER ROT OF APPLES.

BY

HERMANN VON SCHRENK,
SPECIAL AGENT IN CHARGE OF THE MISSISSIPPI VALLEY LABORATORY,

AND

PERLEY SPAULDING, SPECIAL AGENT.

VEGETABLE PATHOLOGICAL AND PHYSIOLOGICAL INVESTIGATIONS.

ISSUED JULY 18, 1903.

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BULLETINS OF THE BUREAU OF PLANT INDUSTRY.

The Bureau of Plant Industry, which was organized July 1, 1901, includes Vegetable Pathological and Physiological Investigations, Botanical Investigations and Experiments, Grass and Forage Plant Investigations, Pomological Investigations, and Experimental Gardens and Grounds, all of which were formerly separate Divisions, and also Seed and Plant Introduction and Distribution, the Arlington Experimental Farm, Tea Culture Investigations, and Domestic Sugar Investigations.

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[Continued on p. 3 of cover.]

APPLES AFFECTED WITH BITTER ROT.

Inoculation from a canker.

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B. T. Galloway, *Chief.*

VEGETABLE PATHOLOGICAL AND PHYSIOLOGICAL INVESTIGATIONS.

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[a] Detailed to Botanical Investigations and Experiments.

LETTER OF TRANSMITTAL.

U. S. Department of Agriculture,
Bureau of Plant Industry, Office of the Chief,
Washington, D. C., April 8, 1903.

Sir: I have the honor to transmit herewith a paper on "The Bitter Rot of Apples," by Dr. Hermann von Schrenk, Special Agent in Charge of the Mississippi Valley Laboratory, and Perley Spaulding, Special Agent, Vegetable Pathological and Physiological Investigations, and respectfully recommend that it be published as Bulletin No. 44 of the series of this Bureau.

This paper was prepared under the direction of and was submitted for publication by the Pathologist and Physiologist. The illustrations, which comprise nine half-tone plates and nine text figures, are an essential and important part of the paper.

Respectfully,

B. T. Galloway,
Chief of Bureau.

Hon. James Wilson,
Secretary of Agriculture.

PREFACE.

For the past four or five years the bitter rot of apples has been the cause of heavy loss to growers and handlers of this fruit. As stated in our report for 1901, the president of the National Apple Shippers' Association estimated that the damage to the apple crop of the United States in 1900 from bitter rot was $10,000,000. In some orchards there was a total loss of fruit; in others from one-half to two-thirds of the crop was destroyed. The disease is especially severe in the Mississippi Valley and the States along the Ohio River. At the request of numerous growers this Bureau undertook extensive investigations to determine more definitely the life history of the fungus causing bitter rot with the hope of discovering a more effective method of holding it in check. The report presented herewith contains a general account of the history of the disease, a description and life history of the fungus causing it, and some facts which have been recently discovered in regard to the mode of life of the parasite.

During the year 1901 cooperative experiments, conducted along lines suggested by this Bureau, were carried on with the Illinois Experiment Station, but during the last season the work was conducted independently by both the station and the Department. Cooperative experiments on the control of this disease were started the past year with the Missouri Fruit Experiment Station, and will be continued with this station and fruit growers in various apple sections during the present season.

ALBERT F. WOODS,
Pathologist and Physiologist.

OFFICE OF THE PATHOLOGIST AND PHYSIOLOGIST,
Washington, D. C., April 7, 1903.

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THE BITTER ROT OF APPLES.

INTRODUCTION.

The bitter rot or ripe rot of apples has for many years formed one of the most serious enemies of this fruit. It made its first appearance in the United States before 1867, according to Curtis's catalogue, but it was not specifically described until 1874, when M. J. Berkeley and M. A. Curtis published the first descriptive notice concerning its occurrence in America. With the increasing number of apple orchards throughout the central belt of States, its range and destructive action have steadily increased.

The bitter rot is a disease of the ripening fruit, which appears late in the summer, affecting whole orchards at once and destroying vast quantities of fruit when it is almost ready for marketing. Estimates of the loss resulting to apple growers from the ravages of the bitter rot in various sections of the country have been made repeatedly.

The bibliography beginning on page 46 of this bulletin gives in full the titles to which the short citations of authorities in the text of this paper may be referred.

A few statements from various sources will show what this pest is capable of doing:

> This orchard that appears so vigorous and healthy is almost worthless. Last year it had at least 1,000 bushels of apples on, and the proprietor did not get a bushel of winter apples. The bitter rot blasts them like the breath of ruin, and the promise of spring ends in disappointment and decay. * * * This orchard was in its prime from the time it was 8 until it was 18 or 19 years old. For ten or eleven years it gave most bounteous returns and produced wagonloads of the finest fruit. It then began to decline. The fruit commenced to speck, and the evil increased until the trees are little more than an incumbrance on the ground. (Murray, 1870.)

An Arkansas man relates his experience as follows:

> The man from whom I purchased my place told me that the Fameuse had always been subject to the rot. For the last three years the disease has steadily increased, so that this year (1887) my old orchard of 75 trees will not yield 25 bushels of sound apples. (Galloway, 1887.)

In 1900 it was estimated that the loss in four counties of Illinois for that season was $1,500,000. (Burrill and Blair, 1902.)

The Pathologist and Physiologist of the Bureau of Plant Industry of the U. S. Department of Agriculture in 1901 says: [a]

> The losses caused by bitter rot in the Middle States often amount to half or three-fourths of the entire fruit crop, single large growers sometimes losing 10,000 barrels of apples. One firm estimated that their losses in 1900 on apples bought in the orchard, in Missouri alone amounted to $20,000 to $30,000, and orchards which in midsummer promised a yield of 25,000 barrels of choice apples produced only about 5,000 barrels of indifferent fruit, owing to this disease. The president of the National Apple Shippers' Association estimated that the damage to the apple crop of the United States in 1900 from bitter rot was $10,000,000.

In some years the destruction was so great as to cause many fruit growers to abandon the business, and instances have been known where men have leased their bearing orchards at $5 per acre for periods of five years, preferring to be assured of that small amount rather than risk getting nothing from their trees because of the work of this fungus.

Older reports make mention of extensive destruction. Galloway reported in 1889 that "in certain places in Virginia, Kentucky, Tennessee, Missouri, and Arkansas our agents report this season a destruction of from 50 to 75 per cent of the crop."

Garman in 1893 stated that bitter rot probably caused more loss to Kentucky fruit growers than any other disease, and statements of this character have appeared from time to time and with increasing frequency in the reports of experiment stations and horticultural societies.

The sudden appearance of the disease at a time when the grower has spent time, money, and energy in producing a large crop, and the almost total destruction of the apples in a few days, causes the disease to be universally feared. It has probably done more to discourage apple growing in many regions than all other troubles, including both fungous and insect diseases combined.

In spite of the universal and destructive appearance of this disease, comparatively little had been accomplished until recently toward preventing or even checking the bitter rot, although its cause was clearly established by Berkeley in 1856, as described more fully hereafter, while other observers have given detailed accounts of spraying experiments. Ever since the experiment stations were established investigations have been conducted looking toward preventive measures. Many papers on the subject have been written, an idea of the number of which can be gathered by referring to the bibliography at the end of this Bulletin.

In the following pages a general account of the disease, a description of the fungus and its life history, and some facts which have recently been discovered in regard to its mode of life, etc., are presented.

[a] Annual Reports, Department of Agriculture, 1901, p. 47.

HISTORICAL ACCOUNT OF THE BITTER ROT.

The early accounts of the bitter rot deal mainly with the fungus causing the disease, which for the present we will call by the name which it has held for so many years—*Glœosporium fructigenum* Berk. Rev. M. J. Berkeley described a fungus causing a ripe rot of grapes in 1854, which was probably the same as the bitter-rot fungus (see fig. 4). Two years later he described a fungus causing ripe rot of the apple, calling it *Glœosporium fructigenum* n. sp. He describes (1856) the disease (see fig. 5) as follows:

> It (the apple) presented a spotted appearance externally as well as internally. * * * The spots were perfectly circular and well defined, and exhibited traces of vegetation. On cutting through the apple the flesh was found to be discolored in various places from the effects of incipient decomposition which was not confined to the surface but penetrated into the center of the fruit. * * * In a few days some of them (the spots) were studded with pearl-like specks bursting through the cuticle and swelling above it in the form of little flat cushions. Sometimes there was but a single speck in the very center, but more frequently there was a more or less perfect ring of satellites, * * * the cuticle was raised into little shining pustules, and a tendril of minute spores * * * was protruded through it.

In 1859 Berkeley described a fungus causing a disease of peaches and nectarines, which he called *Glœosporium laeticolor* n. sp.:

> Nearly a month since we observed on the peaches little dark specks with a bleached center. * * * Two days ago he (the gardener) called our attention to its present condition. The specks were prevalent on the nectarines as well as the peaches. * * * It is of the disease, then, as developed on the nectarines more especially that we are speaking. * * * The white spot and the dark ring around it were most beautifully defined, seated in the center of a regular circular depression, the borders of which were pale, but not completely bleached like the center. The whole surface of the depression was studded with little salmon-colored warts, disposed more or less in circles, from the center of some of which, but especially of those in the bleached cuticle, a little curled tendril of salmon-colored spores was protruded. After a time, however, the several spots run together, and form a depression an inch or more across, still teeming with the red spores.

This fungus is apparently the same as *Glœosporium fructigenum* Berk. Berkeley in his description gives most of his attention to the fungus rather than to the disease caused by it, but we can very easily recognize the fungus of the bitter rot as being the same that he described.

In 1867 Rev. M. A. Curtis in a catalogue of the plants of North Carolina mentions a fungus, *Glœosporium versicolor* n. sp., as occurring upon rotten apples. This was the first use of the name, and also was the first record of the occurrence of a fungus causing bitter rot in this country, as far as can be determined. The bitter rot was very probably known at that time, and possibly quite extensively known, as the article by Murray in the Illinois Horticultural Society publication only three years later (1870) would seem to indicate. In 1874 Berkeley and Curtis published the first description of *Glœosporium versicolor* n. sp., so that the name really dates from 1874.

Practically all of the publications in regard to the bitter rot until 1887 were mere reprints of the articles already mentioned, and the disease was treated apparently as a rather uncommon one and not of much importance.

Galloway seems to have been the first to treat upon the subject of this disease from an economic standpoint, the first accounts dealing with the bitter rot as a destructive orchard disease being published by him in 1887. He called attention to the damage caused and the results of experiments made to check the disease. At that time the bitter rot had appeared in many States from the Atlantic seaboard to Kansas, and the destruction of apple crops was large. These experiments were followed by many others: Garman (1889), Galloway (1889), Jennings (1890), Curtiss (1890), Galloway (1890), Churchill (1890), Chester (1890), Garman (1890), and others. The results of these various experiments were very conflicting. A few investigators succeeded in totally checking the disease even after it had become well established, while others had no success whatever. In the more northern States experimenters seemed to succeed in checking the disease by spraying affected trees with the ordinary fungicides. In the region south of the fortieth parallel, i. e., in the territory extending from the eastern coast to Kansas, Indian Territory, and Texas, where the fungus seems to flourish best, it was found much more difficult to control the disease. Spraying experiments indicated that the disease could be checked to some extent, but only in one or two instances was it stopped entirely. From the time of its first appearance in July until the latter part of September the bitter-rot fungus was active. What became of the spores, where they remained over winter, and how they infected the fruit the following year was unknown, but the opinion was generally accepted that many of the spores survived in the mummified fruits which remained on the trees or on the ground throughout the cold season.

Up to within three years ago it was generally accepted that *Glæosporium fructigenum* Berk. was the cause of the bitter rot of apples. During the last two years, owing to the increased ravages of the disease, attention was directed toward the investigation of the life history of the bitter-rot fungus, with the result that a number of new facts of considerable importance have been determined. These facts have been discovered almost simultaneously by a number of observers, and their exact bearing on preventive measures has already been tested.

DISTRIBUTION OF THE BITTER-ROT FUNGUS.

GEOGRAPHICAL DISTRIBUTION.

The bitter-rot fungus, like other species of the form genus Gloeosporium, has an almost world-wide distribution. In the United States it has been found in nearly all of the States east of and including Kansas, Oklahoma, and Texas. A careful search through the mycological

literature available at the Missouri Botanical Garden has shown that under one name or another this fungus has been reported from Maine, New Hampshire, Vermont, Connecticut, New York, New Jersey, Delaware, Maryland, West Virginia, Virginia, North Carolina, South Carolina, Alabama, Mississippi, Kentucky, Ohio, Indiana, Illinois, Michigan, Wisconsin, Missouri, Arkansas, Kansas, Oklahoma, Indian Territory, and Texas. The States east of the Mississippi from which the fungus has not yet been reported are almost unexplored mycologically. It is extremely probable that it occurs in all States where the apple is being grown, even in the most northern latitudes. (See fig. 1.)

The bitter-rot fungus was first described in England from English

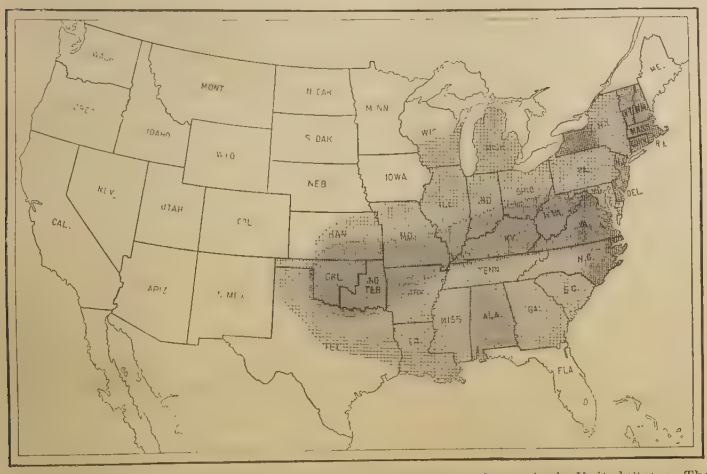

FIG. 1.—Map showing geographical distribution of the bitter-rot fungus in the United States. The shaded sections show where bitter rot occurs, the heavier shading indicating where the disease is most prevalent.

specimens. On one or another host the bitter-rot fungus has been found in all parts of the world. It is reported by Trail (1888), from Perth, Scotland; by Saccardo (1881), from Italy; by McAlpine (1895), from Queensland and New South Wales; by Cooke (1892), from Queensland; by Speschnew (1897), from Tiflis, in Transcaucasia; by Klein (see Frank, 1896), from Baden, Germany; also by Kirchner (1890), from various parts of Germany; by Nypel (1896), from Liege, Belgium; by Viala (1887), from France. It is probable that the fungus occurs to some extent in every country where the apple is grown, which is not surprising when one considers the manner in which apples are sent to all parts of the world and the excellent chances which spores of this fungus have for dissemination with the fruit.

OCCURRENCE ON VARIOUS HOSTS.

The first bitter-rot fungus was described in 1854 (Berkeley) as growing on grapes. Some years later the same writer described a fungus causing ripe rot of the apple. Berkeley in some later papers describes the ripe-rot fungus (under various names) as occurring on grapes, apples, peaches, and nectarines. In 1871 Berkeley, referring to a manuscript description of a fungus growing on grapes (*Glœosporium uvicola*), takes occasion to say that in his opinion it is the same as the fungus causing ripe rot of peaches and nectarines (*Glœosporium læticolor* Berk.). He states that "where grapes are grown in the same house with stone fruits the malady may spread upward," which may be taken to imply that he considered that the ripe-rot fungus of grapes may cause the disease of peaches and nectarines. He further emphasizes their probable identity by saying (p. 1163): "Both on grapes and stone fruits the spores of the fungus are very variable in size, so that no stress can be laid on mere measurement."

Von Thümen (1887) added the pear and the apricot to the list of hosts. Galloway, in 1890, demonstrated for the first time that the ripe-rot fungus of the grape, when transferred to the apple, caused the ripe rot or bitter rot of the apple, and vice versa. Miss Southworth, in 1891, confirmed these results.

Halsted, in 1892, published the results of a large number of experiments in which he had inoculated fruits with spores from different ripe-rotted fruits to determine whether the numerous forms of ripe rot or bitter rot were really due to the same fungus. His experiments seemed to prove that the same fungus caused the ripe rot of the apple, grape, peach, pear, pepper, tomato, and eggplant. The fungus was found to grow on bananas, quinces, lemons, and beans, in addition to the other hosts mentioned.

Chester (1893) corroborated Halsted's results so far as the tomato, grape, pepper, and apple were concerned. He came to the same conclusion as Halsted—i. e., that the fungus on the apple, grape, tomato, and pepper is biologically the same species. Alwood (1894) states that all pomaceous fruits are attacked by *Glœosporium fructigenum* Berk. During the last year Spaulding grew the fungus successfully on squash. (Pl. IV, fig. 6. See also Pl. IV, fig. 5, showing the fungus on the pear.)

From the evidence at hand it appears that this fungus can adapt itself to numerous hosts. It seems probable that all the forms are one and the same species, but it will be necessary to develop the perfect or ascus stage of many of them before any positive statement on this point can be made.

GENERAL DESCRIPTION OF THE BITTER ROT.

TIME OF APPEARANCE.

The bitter rot appears in an apple orchard at different times during the months of July and August, the time of its first appearance varying with the climatic conditions during any particular season. The first spots (Pl. IV, fig. 2) usually develop on the apple fruits when they are nearly full grown. From that time on until the fruit is entirely ripened the disease is likely to occur with increasing severity. In the Southern States bitter rot may destroy some fruit in the early part of July. In a number of cases apples only three-fourths of an inch in diameter were found affected with the disease. One of the writers collected apples affected with bitter rot in Vermont on October 20. The spots were small but well developed, and were present on a large number of individual fruits on one tree.

The factors which determine the time of appearance are probably (1) the age of the fruits; (2) the temperature and humidity of the air; (3) the presence of spore-distributing centers. The age of the fruit is a factor of considerable importance. As a rule, the green fruit is comparatively immune, which may be due in part to the large amount of malic acid present in the unripened fruit. It is possible to produce the disease on green fruits by artificial inoculation by allowing such fruits to lie on a shelf for several days after being picked. Different varieties of apples show a different susceptibility with respect to the time of attack. No hard and fast rule can, however, be laid down in respect to this matter, as the climatic conditions may hasten or retard attack.

Warm, sultry weather, particularly after a rain, forms the ideal condition for the development of the bitter rot. In cool, dry summers the bitter rot is usually present but sparingly. A short series of hot, wet days in August may bring about a sudden and very destructive attack. Nights with a heavy fall of dew alternating with hot days are usually followed by an extensive development of the disease. Numerous instances might be mentioned where the disease appeared in an orchard during the latter part of August, after a few hot days, destroying the whole crop in three days. A notable case of this kind occurred during the summer of 1900. Cold weather usually checks the disease and may stop it altogether.

The time of appearance of this disease is probably influenced also by the condition of ripeness of the spores in the cankers (as described later) and in the mummies. A cold spring may retard their development and consequently bring about a late attack on the fruit, or vice versa.

CHARACTER OF THE SPOTS.

The first signs of the bitter rot appear in the form of a very faint light-brown discoloration under the skin of the apple. The spots are exceedingly small at first, and as they grow larger they appear circular in outline. (Pl. IV, fig. 2.) The spots rapidly increase in size, becoming darker brown. When the spot is one-eighth of an inch in diameter the area appears distinctly sunken. The borders of these spots are usually very nearly circular and sharply defined. When about one-half an inch in diameter small black dots appear at more or less regular intervals beneath the epidermis in the sunken area. These increase in size and project as tiny raised points. At a later stage they break through the epidermis of the fruit and allow large numbers of spores to escape. (Pl. IV, figs 1 and 7.) These spores, when not washed from the fruit, form pink masses, sticky when moist. As the spore mass dries it cakes and adheres to the epidermis. On quiet, dry nights the spores are discharged in long tendril-like threads (Pl. VI, fig. 2), oozing out slowly from the mouths of the black bodies, which are the fruiting bodies of the bitter-rot fungus. These black bodies or pustules are often arranged in the form of a ring. (Pls. I and II.) As the rot progresses other rings of pustules appear outside of the first one, and at regular intervals six to eight, and sometimes more, well-defined rings may form in rapid succession. Each ring will have hundreds of pustules, each producing spores at the same time, so that some rings appear almost continuous. (Pls. I, II, and IV, figs. 1 and 7.) The formation of these rings depends on the rapidity with which the fungus grows. The most perfect rings of pustules are formed when the fungus grows most rapidly. (Pls. I and II.) Cold weather will be followed by a more or less irregular development of the pustules. (Pl. IV, fig. 3.) They then break through the epidermis at many points, as shown on Pl. IV, figs. 3 and 7. The arrangement of the fruiting bodies in rings is a common phenomenon among fungi. Where a single spore germinates in a medium where the food supply is abundant on all sides, the hyphæ generally grow in all directions with equal rapidity. When the period for the development of spores has arrived, the spores will be formed from hyphæ of the same age, i. e., at points equally distant from the original point of infection. The phenomenon of fairy rings is a notable instance among the higher fungi. Alternate periods of low and higher temperature may account for the intermittent development of fruiting bodies, and hence the formation of successive rings.

The pinkish appearance of diseased fruit is due to the spore masses which exude from the pustules. After a rainstorm the interior of the pustules looks sooty black and the mouths ragged, all the spores having been washed off.

The brown coloration of the spots on the apple fruit is an indication of the decayed condition of the tissues immediately under the spots, in which region the threads of the bitter-rot fungus are bringing about changes in the firm tissue of the fruit which make it appear decayed. The lateral progress of the disease, evidenced by the increasing diameter of the brown area, is accompanied by a corresponding progress of the disease into the fruit (figs. 2 and 3). The rotted mass, which is an inch in diameter at the surface, usually extends inward to the core of the fruit. The rotted mass shrinks somewhat in volume, hence the sunken character of the spot outside. There is usually a sharp dividing line between the rotted mass and the sound tissue. In this respect the bitter rot differs from the black rot of the apple.

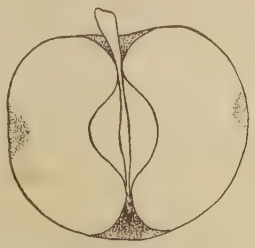

FIG. 2.—Diagram showing how the bitter-rot fungus decays an apple.

The size of the diseased areas on the fruit increases rapidly after an infection, and eventually the whole fruit may be affected. Where two or more separate infections take place the diseased masses fuse (Pl. I). The separate rings of fruiting bodies join and the two sets of hyphæ then grow on just as if there had been but one. The completely rotted fruit appears considerably shrunken, especially if there have been several centers of infection. The fruit hardly ever decays entirely, as do apples attacked by the black rot; as a rule there are small patches of healthy cells which hold out a long time. The affected fruit falls from the tree during all stages of the disease. (See Pl. III.) In this case, as in other instances of fruit diseases due to fungi, a hastening of the ripening process takes place. The diseased fruits are heavier than ripe fruits, and are readily shaken from the trees. There are probably other changes taking place in a diseased fruit which influence its condition and bring on premature fall, much as with fruits stung by various insects.

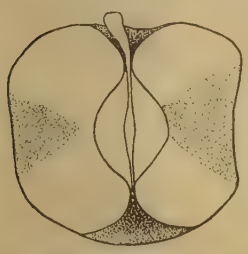

FIG. 3.—A later stage of bitter-rot decay.

CAUSE OF THE BITTER ROT.

The bitter rot of apples is due to a fungus, *Glœosporium fructigenum* Berk., which grows in the ripening tissues of the fruit, thereby inducing decay. The earliest accounts of this fungus deal largely with its systematic position, but it was recognized at an early date that the

bitter rot or ripe rot of both apples and grapes was in some way connected with this fungus. A complete description of the fungus and its various stages is given in a succeeding chapter, and it will be necessary at this point to simply mention in brief the general appearance of the fungus in the orchard and the reasons for connecting this fungus with the bitter rot.

Reference has already been made to the appearance on diseased fruits of more or less regular rings of pustules. In these pustules small one-celled spores are formed, which exude from the pustules after they have broken through the epidermis. In the orchard these spores seem to be produced abundantly only when the air is heavy with moisture. Heavy dews and rain wash away the pink spore masses, leaving the ragged mouths of the pustules freely exposed. The appearance of the pustules and the discharge of the spore masses are the only evidences of fungus activity visible during the attack.

These pustules and spores are, however, always present in apples affected with the bitter rot, and no cases of this disease are known where these spores have not appeared at one stage or another. Moreover, inoculations of sound fruit with the spores of *Glœosporium fructigenum* Berk., made by many experimenters, have invariably produced the disease (Pl. II). There is, therefore, no doubt whatever that the bitter rot or ripe rot of apples and of some other fruits is caused by the bitter-rot fungus (*Glœosporium fructigenum* Berk.).

RATE OF DEVELOPMENT OF THE BITTER ROT.

The rate with which this disease develops depends largely upon conditions of temperature and moisture. During some summers it may take several weeks for the disease to develop to any extent after its first appearance. Then, again, it may start on a fruit during the night and in three to four days entirely destroy it. Apples inoculated with the bitter-rot fungus and kept in incubators at a temperature of 37° C. (98.6° F.) will show spots three-fourths of an inch in diameter, with numerous pustules, in forty-eight hours after inoculation. As has been stated above, the disease will develop in several days in epidemic form when heavy dews fall during the night, followed by hot, cloudy days. The condition of the apple fruit at the time of the attack influences to some extent the rate of development. Thus green fruits do not suffer as severely from the disease nor does the disease progress as rapidly as with fruits which are almost ripe. The disease progresses at different rates on different varieties of apples. Some are more easily affected than others. In general, it may be said that hot, muggy weather is most favorable for the rapid development of the bitter rot.

THE DISEASED APPLE.

An apple affected with the bitter rot or ripe rot is a most objectionable fruit. The name of the disease is derived from the peculiar bitter taste of the decayed tissues of the fruit which is noticed almost as soon as the fungus has begun its growth in the cells. One observer (Alwood, 1894) states that this bitter taste is not always present. The partially decayed fruit leaves a bitter taste in the mouth, resembling the after effect of quinine, yet not quite the same. The bitterness increases as the rot becomes more pronounced. The tissues of the apple are hard and firm when first affected; the cells look somewhat watery and are pale brown. As the rot advances the flesh of the apple softens and turns darker in color, until at an advanced stage the whole tissue is soft and mushy, very watery, and without any resemblance to the original healthy tissue. Attention should be called to the fact that in no case does the apple become as soft and mushy as it does when affected with most other fruit-rotting fungi, for instance, the black rot. The decay starts at the surface of the fruit and gradually extends inward toward the core, making a sort of cone-shaped mass of diseased tissue, as described above (figs. 2 and 3).

During the early stages of the disease there is a marked accumulation of starch around the affected spots, which calls to mind similar phenomena described by Halsted (1898) for various leaf-spot fungi. The cells of the apple tissue separate from one another as the disease progresses. The middle lamella of the cells is dissolved by the fungus hyphæ, but the cell walls themselves remain intact.

THE BITTER-ROT FUNGUS.

LIFE HISTORY ON APPLES.

The spores of the bitter-rot fungus germinate on the apple fruit when it is nearly ripe. In some cases the fungus has attacked apples when they were only three-fourths of an inch in diameter (Garman, 1893), which, however, may be regarded as exceptional. The hyphæ from germinating spores enter the apple and begin to grow in the layer immediately under the epidermis. Whether the young hyphæ can pierce the uninjured epidermis of the apple seems to be a somewhat disputed point. In making infections of apple fruits in the laboratory it was found that the greatest numbers of successful infections were obtained by puncturing the epidermis with a sterile needle and then spraying the spores on to the broken epidermis. Clinton (1902) states that the spores placed on unpunctured apples, "if successful," brought about the rot two or three days later than when placed on punctured fruits. It is probably true that the young hyphæ can enter through the unbroken skin, possibly through the stomates, but at the

same time it is probable that a large percentage of the infections in an orchard start in fruits which have been wounded in some way, generally by insects. When one reflects that the number of spores which fall on a fruit is generally very large, it is strange that there should be only very few infections or sometimes only one infection. This point is one which will require additional careful study.

After the first hypha has entered the tissue below the epidermis it branches rapidly. The hyphæ grow in the intercellular spaces, absorbing the sugar and other products from the apple cells. (See Pl. V, fig. 3.) The affected cells turn brown and separate, and after a time they collapse. It is then that the presence of the fungus becomes noticeable on the outside in the form of the brown, sunken spots mentioned above (Pl. IV, figs. 1 and 2). The fungus hyphæ grow in all directions from the original point of infection with great regularity. As they extend outward the cell groups attacked become brown in turn and collapse more or less. This regular development gives the affected mass of cells the circular form visible on the outside.

Early in the season the brown areas are about one-half of an inch in diameter before there is any evidence of spore formation; later on, during the height of an epidemic, the spores begin to form when the affected areas are still very small. The spore-forming stage is evidenced by the appearance of numerous small raised points, which push up the epidermis in a brown spot at irregular intervals. These points are composed of masses of parallel hyphæ which grow outward from the cells just underlying the epidermis. These hyphæ are short and so arranged as to form a low cone, whose apex pushes against the epidermis as the hyphæ composing it grow in length. These hyphæ are at first colorless and then turn olive colored. Ultimately, either by pressure or because of the solvent action of an enzyme, the tip of this cone breaks through the epidermis. On the outside the tips of the cones appear as small dark specks. The unicellular spores are formed by abstriction from the ends of the hyphæ composing the cone, many spores being formed from each hypha. Great masses of these spores issue from the hole made in the epidermis and remain on the outside as bright pinkish, glistening masses, adhering to the tips of the cones. The latter are the pustules or sori. When the fungus is growing rapidly the pustules or sori may form when the spots are but one-fourth of an inch in diameter. The spore masses are sticky and adhere firmly to the mouth of the pustules. Sometimes, especially during nights when a heavy dew has fallen and there is an abundance of moisture in the air, the spores will be discharged forcibly in the form of tendril-like masses. (Pl. VI, fig. 2.)

As stated above, the sori appear in irregular groups early in the season. Later in the summer, when the fungus is growing rapidly, they break through the epidermis in groups, forming very regular rings (Pl. I). These rings are striking objects shortly after the dis-

charge of the spores and form one of the most characteristic features of the disease. The spores disappear from the tip of the pustules after a time. Rain or dew may wash them away or insects rub them off. The empty sori remain behind and have a sooty, black appearance.

The time elapsing between spore germination on a fruit and the ripening of the first spore crop differs with the season from three or four days to a week. In hot days of August the cycle is completed with great rapidity. In one and the same spot on a fruit spores may be forming at the center, while a quarter of an inch farther out the pustules have not yet begun to develop.

The foregoing description of the growth of the fungus pertains to the development on apples still on the trees. Spores inoculated into apples after they have been picked will give rise to similar phenomena. The rate of growth of the fungus and the formation of spores will depend entirely on the temperature and moisture conditions under which the inoculated apple is kept.

The spores of *Glœosporium fructigenum* Berk. germinate on grapes when they are almost or quite ripe (Southworth, 1891). The bitter taste which follows the attack of this fungus on apples is absent in diseased grapes. Hence the more common designation of "ripe rot" for the same disease of the grape. On white grapes small reddish-brown spots appear, which spread and become darker as they grow older, until the spots have an almost purple center with a bright brown border. The pustules on the grape are at first white, then darker, until they are almost black. The spores are flesh-colored. The berry ultimately dries up, but does not turn black. Dark-colored grapes show no color changes when attacked.

THE CONIDIA

The spores of *Glœosporium fructigenum* Berk. produced in the sori, and commonly called conidia, are pinkish-colored en masse. This color varies from a light fresh pink to a darker reddish pink.[a] When highly magnified they have a very delicate light-green color. This color is quite distinct, and it seems strange that of many observers Alwood (1894) seems to be the only one to recognize this greenish color. Clinton (1902) states that the conidia are colorless, while Miss Southworth (1891) says that they are hyaline.

In size and form the conidia are extremely variable. The great variability in these respects has probably been responsible for the difficulties which many observers have labored under when it came to deciding which of the several fungi causing similar diseases of fruits

[a] One of the writers noted apples which had been inoculated in the laboratory and bore only cream-colored spore masses. The spores seemed to be perfectly normal in other ways.

properly belonged to this particular species. Alwood (1894) was able to produce in the same culture spherical, dumb-bell-shaped, oblong, ovoid, and cylindrical conidia. Although such extreme forms were not found by the present writers, they agree with Alwood and others that the form variation is certainly large. The general form of the conidia developed on fruits and in cultures may be characterized as oblong or cylindrical, sometimes slightly curved (Pl. V, fig. 1).

Extremes in sizes, gathered from all other writers, were from 6 to 40 μ in length and from 3.5 to 7 μ in width. The dimensions given by some may be mentioned. Alwood gives 10 to 12 $\mu \times 4$ to 6 μ. Saccardo's measurements, 20 to 30 $\mu \times 5$ to 6 μ, are probably of exceptionally large spores. The average size, as determined by the writers, is 12 to 16 $\mu \times 4$ to 6 μ.

Miss Southworth says of the conidia: "They are apt to be shorter and thicker on the apple [than in cultures], and in dry than in moist surroundings."

The spores can not and should not be taken as a criterion in determining whether any particular fungus is *Gloeosporium fructigenum* Berk., since the spores of other species of Gloeosporium closely resemble those of the bitter rot. The great variability in size and form of many fungi of a more or less saprophytic nature is coming to be more widely recognized, and the former method of lumping or separating many forms simply by spore characteristics is rapidly giving way to a clearer conception of the relationship based upon more constant characters.

The ripe conidia are filled with a finely granular protoplasm. Near the middle and usually a little to one side a clear hyaline area is generally visible (Pl. V, fig. 1). It is at this point that the septum forms during germination. Normally, the conidia are one-celled until they germinate. They resemble the ascospores of this same fungus, and the two can hardly be distinguished. As a rule the ascospores are slightly curved, while the conidia are straight.

GROWTH IN CULTURES—CONIDIAL AND ASCUS STAGES.

Freshly-formed conidia of the bitter-rot fungus germinate in three or four hours when put in water at room temperature. Just before germination a septum frequently forms at or near the middle of the spore, thus making a two-celled spore (Pl. V, fig. 2). A spore may produce one, two, or more rarely three, germ tubes (Pl. V, fig. 2). Where a wall forms, these two germ tubes start, one from each end of the spore. Short spores generally have but one tube. When the spores germinate in drops of water they become vacuolated after a few hours, and after five or six hours they become entirely empty. When germinated in bouillon or on agar, the protoplasm remains finely granular for some time and the cells rarely become entirely emptied.

The germ tubes grow in length with great rapidity, reaching a length three or four times that of the spore (Pl. V, fig. 2) in three-fourths of an hour. In a water medium the first hyphæ grow to considerable length before branching. In bouillon or agar they branch when two to three times as long as the spore. Septa form very early in the development of the mycelium (Pl. V, fig. 2).

Fusions between neighboring hyphæ are common both in the apple fruit and in cultures. The young hyphæ are colorless and are filled with a granular protoplasm. When growing in the tissues of the apple, the young hyphæ soon turn darker and ultimately become brown. When an abundant food supply is at hand the mycelium grows to large dimensions, and it may be several days before any fruiting bodies are formed. These are usually conidia formed by a process of abstriction at the end of short lateral hyphæ (Pl. V, fig. 7).

These conidia develop with great rapidity (Clinton, 1902), so rapidly that in twelve hours an agar plate will appear as if covered with a powdery mass. When growing under unfavorable conditions, so that the mycelium is starved, some of the hyphal tips will swell considerably, and a wall will cut off the swollen end (Pl. V, fig. 2). The walls of this swelling turn dark red-brown and thicken somewhat, forming what appears to be a spore (Pl. V, fig. 2). A bright translucent spot is usually present near the center. These brown bodies have various shapes and appear to be formed by most species of the genus Gloeosporium. Miss Stoneman (1898) figures them for *G. fructigenum* and *G. naviculisporium*. Miss Southworth (1891) and Clinton (1902) obtained them in cultures of *Gloeosporium fructigenum*. *Gloeosporium cactorum* forms very fantastic bodies which bud and develop short tubes (Pl. V, fig. 4). Halsted (1892) published an extended account dealing with some of these secondary spores. Many attempts were made to cause the bodies formed by *Gloeosporium fructigenum* to germinate, but so far without success. They probably represent a form of chlamydospore, which may have to undergo a resting period before developing.

The conidia formed freely from rapidly growing mycelium on agar resemble those from the pustules on apples in all respects. They germinate in a similar manner, and the mycelium which they give rise to may produce similar conidia, pustules, or perithecia, as the case may be, depending upon the age of the fungus, the food supply, etc. The fungus can be made to grow continuously, producing crops of conidia without the production of the other stages.

When kept growing on apple agar several crops of conidia usually form, as described above, and when the food supply has been partially exhausted the production of conidia gradually stops. The first lot of conidia have germinated by this time and have produced mycelia, so that a petri dish with a pure culture of the bitter-rot fungus

is covered with a dense growth of mycelium after a period of from ten to fourteen days. At about the time when the conidia cease to be formed small black knots appear among the tangled mass of hyphæ, looking much like warts. Drops of a yellowish liquid frequently exude from these black bodies (Pl. VI, fig. 1) The latter increase in size and frequently form masses one-fourth of an inch in diameter. Cultures on apple agar will show good-sized masses of this kind in from twelve to eighteen days. These black masses contain the perithecia of the bitter-rot fungus.

The perithecia and asci were first described by Clinton (1902), who proved their connection with the bitter-rot fungus (*Glæosporium fructigenum* Berk.) by inoculating ascospores into apples and producing the bitter rot. The formation of perithecia was found by the writers to occur with great constancy under appropriate conditions. As a rule the perithecia form in older cultures only.

The black nodules in which the perithecia are embedded are hard masses of mycelium, which may be characterized as carbonaceous. They are very irregular in shape and vary in size from a small pin head to one-fourth of an inch in diameter. The perithecia, from one to many, are embedded in this carbonized mass. In apple-agar cultures the perithecia form when the black nodules are still very small. When there is but one perithecium in a nodule it is almost spherical; when there are several, they are somewhat flattened laterally, and sometimes very irregular in form. There is no beak. Clinton (1902) found the perithecia to be from 125 to 250 μ in length. These measurements agree fairly well with those found by the writers. The walls of the perithecia show marked reticulations about 6 to 14 μ in diameter. These are quite marked in the early stages, but become obscured as the perithecium matures.

The asci (Pl. V, fig. 6), which occur in considerable numbers in a perithecium, are oblong-clavate in form, 55 to 70 μ by 9 μ, often with a slight pedicel, and are comparatively thin walled. When mature they break open and disappear rapidly. They contain 8 ascospores, which are usually arranged in pairs, more rarely in oblique series. The spores resemble the conidia formed directly on the mycelium, so much so that they might easily be taken for conidia. They are perhaps curved a little more than the conidia, a character which can sometimes be used to separate the two, but not always. The ascospores and the conidia are about the same size, though the former are not as variable as the conidia, measuring 12 to 22 μ by 3.5 to 5 μ. Their great resemblance has probably led to the ascospores remaining undiscovered for so many years. The asci are short lived and after they have discharged the spores they vanish. When found in apples it is practically impossible to tell whether any particular spore is an ascospore or a conidium. The ascospores germinate much in the same

manner as the conidia, and the description given for the germination of the conidia will hold for the ascospores.

Conidia were found in the bitter-rot canker during the summer of 1902, and in the latter part of 1902 perithecia and asci were found in the canker. (Science, **17**: 188, 1903.) The description of these will be given below in discussing the cankers.

The bitter-rot fungus grows readily on most culture media. It grows vigorously on apple agar, on sterilized apple wood or leaves, on sterile pine blocks, bean stems, etc. It is in many respects a true saprophyte. It is questionable whether it ought to be considered a parasite at all times when growing on the ripe fruit in the orchard, for at the time it attacks such fruit the latter is practically full grown and is no longer composed of cells or tissues which will react when stimulated. The fungus develops best at temperatures ranging from 33° to 38° C. (91.4° to 100.4° F.). Apples which were kept in incubators at 38° C. after infection showed decayed spots 1 inch in diameter in from one to three days. Cold checks growth materially, and at 2° C. or 35.6° F. (cold-storage temperature) no further growth takes place.

THE NAME OF THE BITTER-ROT FUNGUS.

About the middle of the last century a number of fungi were described by M. J. Berkeley as growing on various fruits and bringing about their decay. In 1854 he published the discovery of a fungus growing on grapes which caused ripe rot. He says of this:

> The surface of the spots is rough, with little, raised, orbicular, reddish bodies arranged in concentric circles and easily separating from the matrix, which is perforated for their protrusion. The outer surface of these bodies consists of delicate cells, with a distinct darker nucleus, and when this is removed a lobed hymenium is seen within, rough, with distinct sporophores, each of which is surmounted by an oblong spore, sometimes constricted in the center, and occasionally so much so as to become pyriform, and varying in size from $\frac{1}{1200}$ to $\frac{1}{800}$ inch. In age the perithecia fall away, leaving a little aperture, the border of which is often stained with black.

Berkeley named this fungus *Septoria rufo-maculans*, n. sp. He figures pycnidia and spores, and for reference the latter are reproduced herewith (fig. 4). In 1860 he changed the name to *Ascochyta rufo-maculans*. Von Thümen (1879) renamed this grape fungus *Glœosporium rufo-maculans* (Berk.) v. Th.

In 1856 Berkeley described a fruit-rotting fungus growing on apples, which he called *Glœosporium fructigenum*, n. sp. This is the first authentic description of a fungus causing bitter rot, or ripe rot, of apples. Berkeley says of this fungus:

> On examination each plant was found to consist of a branched inosculating mycelium, giving rise to simple or forked subfastigiate irregular threads, each tip of which was surmounted by an oblong, curved, or irregular spore about $\frac{1}{100}$ of an inch in length. There was not the slightest trace of an investing membrane or perithecium.

He refers to the description of a grape-rotting fungus two years before, and seems inclined to doubt the wisdom of considering the apple fungus a new species.

> The spores (of the apple fungus) are more inclined to be curved, rather longer, and not so variable in size, and the want of a perithecium separated the two widely from each other. * * * At the same time these organisms are so different in different conditions that I would not affirm that the two productions are essentially different, and the more especially because in external appearance and habit they are so perfectly identical.

Berkeley's figures of this fungus are reproduced in figure 5.

In 1859 Berkeley published the name *Glœosporium lœticolor* n. sp., applying it to a fungus growing on peaches and nectarines. He evidently regarded this species as quite distinct from the apple and grape

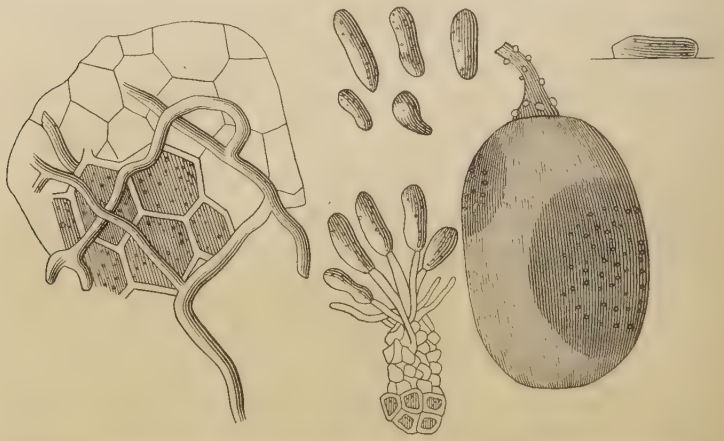

FIG. 4.—Berkeley's grape-rot fungus (*Septoria rufo-maculans* Berk.). [Drawn from the original figure.]

fungi, as he speaks of these in the following words (p. 676): "A plant of the same genus, destructive to apples, is figured and described in this journal (Gardeners' Chronicle, 1856, p. 245). * * * We may also refer to the very similar production on grapes."

In 1874 Berkeley and Curtis described a fungus growing on apples in South Carolina, calling it *Glœosporium versicolor* n. sp. They appeared anxious to emphasize the fact that this new fungus was not *Glœosporium fructigenum*, as they say: "It is very different in habit."

In the years following this last description the accounts dealing with the bitter-rot fungus on apples in the United States speak of it as *Glœosporium fructigenum* Berk., using the name given for the fungus on apples by Berkeley in 1856.

When Miss Southworth, in 1891, published an article on the bitter-rot fungus she reviewed the older accounts of fungi causing bitter rot

or ripe rot of fruits, and decided to accept the name *Glœosporium fructigenum* Berk. for the fungus causing the bitter rot of apples. She gives her reason for so doing in the following words:

> The strict law of priority might demand that we now make the specific name *rufo-maculans*, but since the better-known *G. fructigenum* is also Berkeley's name it will remain so in this paper.

At this point it will be necessary to refer again to the results which Miss Southworth obtained by inoculating grapes with spores of the apple bitter-rot fungus (*Glœosporium fructigenum* Berk.) and apples with the grape ripe-rot fungus (*Glœosporium rufo-maculans* (Berk.) v. Th.). In both cases a bitter rot or a ripe rot of the respective fruits followed, which led Miss Southworth to regard the bitter-rot fungus and the ripe-rot fungus as one and the same species. The experiments of Halsted (1892) seemed to verify Miss Southworth's experiments

FIG. 5.—Berkeley's apple-rot fungus (*Glœosporium fructigenum* Berk.). [Drawn from the original figure.]

and to show that the same fungus caused the ripe rot not only of apples and grapes, but also of quinces, pears, peaches, nectarines, peppers, and other fruits.

Summing up the foregoing, it appears that several fungi causing fruit rots have been described under the impression that they were distinct species. More recent investigations have demonstrated that in all probability the same fungus has caused the various ripe rots of fruits. The different forms variously described as *Glœosporium fructigenum*, *Glœosporium rufo-maculans*, *Glœosporium versicolor*, and *Glœosporium læticolor* probably differ only in minor characters, such as in the size and form of the spores. The effects which they produce on different fruits vary as to color, size of spots, etc. These differences are readily intelligible in view of the better knowledge which we now

have concerning the influence of different substrata on morphological characters of plants. This is particularly true of saprophytic fungi, which, like the ripe-rot or bitter-rot fungus, can grow on media of widely different chemical compositions. An abundant food supply may result in the production of very large spores, just as a meager supply may be followed by the formation of smaller spores.

The various fruits under consideration have a different structure and they differ chemically. It might, therefore, be expected that there would be slight differences in the structure of the fungus, and also in the external appearance of diseased fruits.

In cultures the various forms behave similarly. No such distinct physiological strains as were described for *Neocosmospora vasinfecta* by Erwin F. Smith (1899, p. 39) could be established. This is probably due to the more or less saprophytic nature of the fungus. *Glœosporium fructigenum* Berk. is not bound so strictly to the apple cell as the root-rot fungus is bound to the cowpea, the cotton, or the watermelon.

Assuming, then, that the four species mentioned above are one and the same, it becomes necessary to choose one of the four specific names. According to the generally accepted rules of nomenclature, the name under which the fungus was first known takes precedence, and in this case it is *Glœosporium rufo-maculans* (Berk.) v. Thümen.

This would have been the name of the bitter-rot fungus but for the discovery of the perfect or ascus stage. In 1902 Clinton placed the bitter-rot fungus in the genus *Gnomoniopsis* Stoneman.

In 1895 Miss Stoneman described a new genus *Gnomoniopsis* in which she placed (after obtaining in cultures the ascosporic stage) the following fungi, hitherto known only in the imperfect stages as members of the Melanconiaceæ: *Glœosporium cingulata* Atk., *Glœosporium piperatum* E. & E., *Colletotrichum cinctum* Berk. & Curt., and *Colletotrichum rubicolum* E. & E. Clinton included the bitter-rot fungus *Glœosporium fructigenum* Berk. in this group and named it accordingly *Gnomoniopsis fructigenum* (Berk.) Clinton. It is under this name that the fungus now stands.

In establishing the genus Gnomoniopsis Miss Stoneman, however, overlooked the fact that six years prior to her publication Berlese (1892) used the name *Gnomoniopsis* for a group of fungi very different from the perfect form of the *Glœosporium* which she described. Clinton, in accepting Miss Stoneman's name, likewise overlooked the name published by Berlese. The latter raised the subgenus *Gnomoniopsis* of the genus *Gnomonia* as used by Winter (1887) to generic rank, making the species *Gnomonia chamaemori* (Fries) the type, and including in the new genus the former *Gnomonia miselta* as a variety of *Gnomoniopsis chamaemori* (Fries). Lindau (1902) accepts Miss Stoneman's genus *Gnomoniopsis*. The generic use of *Gnomoniopsis* by Berlese in 1892 clearly invalidates Miss Stoneman's name, and it becomes

necessary to rename the genus. The name *Glomerella* is suggested by the writers. The bitter-rot fungus would accordingly become *Glomerella rufomaculans* (Berk.) Spaulding & von Schrenk, with the following synonymy:

Glomerella rufomaculans (Berk.) Spaulding & von Schrenk.[a]
Septoria rufo-maculans, Berk. (1854, Gard. Chronicle, p. 676).
Ascochyta rufo-maculans, Berk. (1860, Outlines of British Fungology, p. 320).
Glœosporium rufo-maculans, (Berk.) v. Thümen (1879, Fungi Pomicoli, p. 59).
Glœosporium fructigenum, Berk. (1856, Gard. Chronicle, p. 245).
Glœosporium lacticolor, Berk. (1859, Gard. Chronicle, p. 604).
Glœosporium versicolor, Berk. and Curt. (1874, Grevillea **3**: 13).
Gnomoniopsis fructigena, (Berk.) Clinton (1902, Bull. Ill. Agr. Exp. Sta.).

The new genus *Glomerella* stands practically for the genus *Gnomoniopsis* Stoneman, and accordingly includes all the species, four in number, which were placed by her in the genus *Gnomoniopsis*, i. e., they become *Glomerella cingulata* (Atk.) Spaulding & von Schrenk, *Glomerella piperata* (E. & E.) Spaulding & von Schrenk, *Glomerella cincta* (Berk. and Curt.) Spaulding & von Schrenk, *Glomerella rubicolor* (E. & E.) Spaulding & von Schrenk.

Glomerella rufomaculans may be described, using the description given by Clinton (1902), which agrees with the finding of the writers so well that it fully covers all points.

Glomerella n. n. (*Gnomoniopsis* Stoneman, not Berlese). Perithecia membranaceous, dark brown, spherical to flask-shaped, often rostrate, sometimes evidently hairy, cæspitose or more or less compound and immersed in a stroma with which they often form an evident hard cushion; asci oblong to clavate, often fugacious, aparaphysate; ascospores hyaline, apparently eight, distichous, oblong, usually slightly curved, unicellular. Permanent stage of Glœsporium-like fungi.

Glomerella rufomaculans (Berk.) Spaulding & von Schrenk. Permanent stage developing on decayed pomaceous fruits; forming stromatic cushions (often concealed by dark olive mycelial felt), which contain immersed and more or less compounded, subspherical perithecia; asci subclavate, often slightly pedicellate, fugacious 55–70 μ in length, ascospores alantoid, with evident central hyaline area chiefly 12–22 μ by 3.5–5 μ. Glœsporium stage causing rotting of pomaceous fruits; sori small, developing more or less in concentric circles, usually soon rupturing and oozing out spores in small pinkish masses; spores greenish,[b] chiefly oblong, unicellular, with evident hyaline areas when fresh, 10–28 μ by 3.5–7 μ, but chiefly 12–16 μ by 4–5 μ.

To this must be added: The fungus forms cankers on apple limbs, bearing both conidia and perithecia.

THE CANKER STAGE.

DISCOVERY OF THE CANKER.

The sudden appearance of the bitter-rot fungus late in the summer and its equally sudden disappearance in the early winter have long excited conjecture as to where the spores which affected the first apples every year came from. Diseased apples of a previous year hanging

[a] The writers have dropped the hyphen from rufo-maculans in order to simplify the name, as was done by Cooke (1885).

[b] Changed by the writers. The original says "hyaline."

in mummified condition on the trees, or lying on the ground under the trees, probably served as infection centers in many instances; but in many cases, although all mummies and diseased apples were carefully removed during the winter, the disease reappeared in the orchard. Another feature explained with difficulty until recently was the fact that even with many specimens of bitter-rotted apples of the previous season lying on the ground under the trees, the disease first manifested itself in the tops of the trees and very rarely on the branches nearest the ground. In other words, it was difficult to understand how the spores of the bitter-rot fungus got on the fruits in the tree tops from the mummies on the ground without first infecting those on the lower branches. It had been noted repeatedly that the disease frequently made its first appearance on the apple tree in a cone-shaped area, with the apex of the cone near the top of the tree. It was this observation oft repeated which led to the discovery during the past summer of what is probably the winter stage of the bitter-rot fungus.

On July 10, 1902,[a] Mr. R. H. Simpson discovered peculiar depressions on many branches of apple trees in his orchard at Parkersburg, Ill. Mr. Simpson was at that time employed as an agent of the Department of Agriculture to conduct spraying experiments looking toward the control of the bitter rot of apples by spraying with fungicides. Mr. Simpson had been hunting for the source of the first infection, and early in July he noted the peculiar cone-shaped distribution of the fruit which showed the first signs of the bitter rot. On many trees the grouping of the infected fruit in the cone shape was so marked that it seemed probable that the disease had started near the apex of the cone and had spread downward and outward. In nearly every instance Mr. Simpson found blackened depressions of a characteristic appearance on one or more branches at or near the apex of the cone of infected fruit. These black depressions in the apple limbs occurred so constantly associated with early bitter-rot infection that Mr. Simpson proceeded at once to cut out all blackened areas which he could detect. The blackened sunken areas in the apple limbs have the appearance of "cankers," as this term is generally understood, and they have been called cankers since their first discovery. Mr. Simpson was able to locate the canker in more than 95 per cent of the cases by following up the cone of infected fruit to the apex.

On the day following Mr. Simpson's discovery at Parkersburg, Professors Burrill and Blair, of the University of Illinois, visited the orchard at Parkersburg and learned of Mr. Simpson's find. Believing that the causal relation between the cankers and the bitter rot was thereby established, they published a preliminary note in a circular of the Agricultural Experiment Station of Illinois, in which they

[a] The discovery of the apple cankers was made July 10, 1902, in the afternoon, as indicated by a telegram from Mr. Simpson to the writers on the same day, not July 11, as stated in Circular No. 58 of the Illinois Agricultural Experiment Station, July, 1902.

recommended cutting out all cankers in apple orchards. This preliminary circular was followed by a bulletin on the same subject, giving illustrations of the cankers and results of experiments, showing that bitter-rot spores occurred in the canker and that apples could be infected from cankers.

Investigations as to the relations of the cankers and the bitter rot were begun by the writers two days after Mr. Simpson's discovery. These have been continued up to the present time and will be carried on further.

DESCRIPTION OF THE CANKER STAGE.

The cankers found on apple trees in Illinois appear as blackened depressions on apple limbs of various sizes, from last year's fruit spurs to limbs 3 to 4 inches in diameter. Thus far the cankers have not been found on the main trunk. On these limbs rounded or oblong sooty-black sunken spots occur from one to several inches long, which have more or less ragged edges. (See Pls. VII and VIII.)

The entire bark is killed for a considerable distance back (Pls. VII and VIII), and the dead bark appears cracked and fissured and in some instances broken away. In many cankers regular transverse cracks, caused by the drying out of the bark, are very marked. As the bark dries out it adheres very firmly to the underlying wood. As a result of the decrease in volume of the affected bark and cambium, a marked flattening and final depression take place on the affected limb. Around the dead areas a healing callous layer usually forms (Pl. VII, fig. 1; Pl. IX, figs. 1 and 4). This starts at the edges of the dead areas and pushes toward the center, frequently lifting the dead bark at the edges. The appearance of this callous layer makes the cankered spots look more and more sunken. It will be noted that most of the cankered spots show the presence, near the center, of a small branch or of a branch stub. There may be some relationship between the formation of the cankered spot and a diseased fruit borne on such a small branch in a previous year. That is, however, a mere conjecture.

On cross sections of cankers one frequently finds that at its very center the wood has been dead for two years. (See Pl. VIII, fig. 1.) The small hole in the wood, two rings in, shows where the small branch broke away. This dying and breaking away of the small branch would point to the fact just mentioned, that the canker may sometimes start in the branch.

The wood of the branch immediately below a cankered spot is discolored for a considerable distance toward the center. (See Pl. VIII, fig. 1.) The discoloration is brown and resembles that found in many hardwood trees in the region below a wound. The wood cells and medullary rays in the discolored region are filled with a light brown mass, readily soluble in alkalis, which leads one to class it as one of

the humus compounds. It is probably one of the decomposition products which forms when the bark and cambium are killed and which infiltrates the wood. One finds numerous fungous hyphæ in the medullary ray cells and the larger vessels, but at this stage it is not possible to say whether these are hyphæ of the bitter-rot fungus. Further studies in this direction are being made.

The formation of the cankered spot probably starts at some small wound (or branch, as stated above). The fungus begins to grow in the living bark and kills the bark and the cambium. As a result no new wood is formed at the point where the cambium is killed (see Pl. VIII, fig. 1), and a small depression forms as the wood at the edges of the dead cambium increases in thickness. As the fungus grows out from the original point of infection, more and more bark and cambium are killed, until at the end of the growing season a large spot on the limb is dead.

Since there is always a small series of wood cells formed at the beginning of the year during which the attack takes place, the fungus probably starts to grow in the bark early in June. (See fig. 6.)

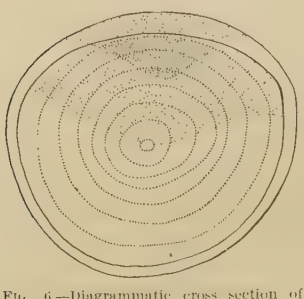

FIG. 6.—Diagrammatic cross section of apple canker.

The majority of the cankers found during the last summer probably were started two years ago. During the first year the fungus made very little headway. A very small central area was killed, generally around and including a small branch. The following year the larger part of the canker was formed. Whether the cankers will continue to increase in size is as yet undetermined, but it does not seem probable, for if such were the case cankers three or more years old ought to have been secured in the orchards where the bitter rot has been common for many years.

RELATION OF THE CANKERS TO THE BITTER ROT.

The discovery of the cankers was brought about directly by tracing groups of diseased apples to these sunken areas on apple limbs. The numerous observations made by Mr. Simpson and by those who followed him seemed to prove beyond question that the cankers were in some way responsible for the infection of the apples. Instances were frequent where two or more apples hung just below a canker. These were generally badly diseased, while all other apples in their immediate vicinity were perfectly healthy.

Although it seemed extremely probable from Mr. Simpson's observations, confirmed and extended by the writers, in his orchard and

in other orchards, that a causal relation existed between the canker on apple limbs and the bitter rot, it was by no means positively proved that the bitter-rot fungus (*Glomerella rufomaculans* (Berk.) Spaulding & von Schrenk) produced the cankers on apple limbs. Experiments were accordingly started to determine whether any such relation existed. Examination of the cankers showed the presence of unicellular spores resembling the spores of *Glomerella rufomaculans* Berk. In most cases there also occurred numerous unicellular brown spores of a fungus which was probably *Sphæropsis malorum* Peck. There were spores present now and then of *Tricothecium roseum* and a species of Alternaria, but the unicellular colorless spores (*Glomerella rufomaculans*) and the unicellular brown spores (*Sphæropsis malorum*) were quite constantly present. The mere presence of spores of any one fungus, even when constantly associated with a canker, is no proof that the fungus producing these spores causes the cankers. It is strong presumptive evidence, but no more. That the colorless one-celled spores were spores of *Glomerella rufomaculans* was proved after a few days by inoculating some of these spores obtained from a canker into healthy apples. These showed unmistakable signs of the bitter rot in a few days. (See Pl. II.) This experiment was repeated many times, using control fruits with every culture. In every case the bitter rot appeared in inoculated fruits, while the check fruits remained sound. Fearing that the spores which caused the disease in these cases might have simply rested in the bark of the cankers, numerous cultures on apple agar were made from pustules in the bark of cankers, and from these pure cultures of the bitter-rot fungus were obtained.

Spores from such pure cultures were inoculated into sound apples, using control fruits, and these also produced the disease (Pl. VI, figs. 3 and 4). These cultures, repeated for several months and under different conditions, left little doubt that the cankers on apple limbs contained spores of the bitter-rot fungus (*Glomerella rufomaculans* Berk.).

A number of tests were made to determine whether the spores could be washed from a canker onto apples by water falling on the cankers. The first test of this kind was made by Mr. Simpson. To insure rapid action on the part of the fungus, he punctured an apple, and then allowed water to run from a canker on the fruit. After several days this apple showed unmistakable signs of the disease.

It now became a matter of considerable importance to determine what connection, if any, existed between the bitter-rot fungus and the cankers. It was very possible that the cankers served merely as lodging places for the bitter-rot fungus or its spores. The presence of numerous spores of what was believed to be *Sphæropsis malorum* suggested that this fungus, which is known to form cankers on apple limbs (Paddock, 1899 and 1900) resembling those in the Illinois

orchards, might be the canker-forming fungus. This supposition was strengthened by the fact that many of the Illinois cankers had the sooty black appearance characteristic of the black-rot apple cankers.

To determine whether the bitter-rot fungus (*Glomerella rufomaculans*) could form cankers, a number of trees in the Missouri Botanical Garden were selected. Small longitudinal slits were cut into the bark, reaching the cambium layer, two slits on every branch. Into the upper slit spores from pure cultures of *Glomerella rufomaculans* (made from diseased apples and from cankers) were introduced. The second slit, from 3 to 5 inches below the first, was used as a control. A large number of control slits were used, as it was possible for spores flying about in the air to enter the infected slits and thereby vitiate the results. It may be said at this point that in no case did any of the control slits show any signs of canker formation. A number of inoculations were made, using pure cultures of *Glomerella rufomaculans* obtained from apple cankers in Illinois and from diseased apples. Inoculations were likewise made with ascospores obtained in apple agar cultures. The first infection of apple limbs was made July 16, 1902.

The inoculations were made by inserting a needle with spores into the freshly made slit, or by spraying water into the slit and then placing some spores in the drop of water. Some of the slits were covered with grafting wax or with cloth waxed with a cocoa-butter mixture, as it was thought that the uncovered slits might be infected by bitter-rot spores from the air. The results showed that this precaution was useless, as none of the uncovered control slits showed any signs of being infected.

Several weeks elapsed before there was any evidence of development on the limbs. In both the inoculated slits and the control slits the bark dried somewhat along the edges of the slit, making a gaping wound. After some two weeks a distinct callous layer had formed under the edges of the bark of the control slits. The two callous layers joined after six to eight weeks and occluded the wound. In the slits where bitter-rot spores had been inserted the callous formation was less marked. The exposed wood turned dark, almost black, and the exposed edges of the bark turned back. The living bark then began to dry out gradually and became depressed (Pl. IX, fig. 3), and after about two months a decided sharply defined depressed area had formed, with the slit in the center. Shortly thereafter small black pustules broke through the dried bark in a number of instances (Pl. IX, figs. 2, 4, 5, and 6). By that time the infected points showed all the characteristics of small cankers.

On examination the black pustules were found to contain masses of spores resembling those of the bitter-rot fungus (*Glomerella rufo-*

maculans). At this time there were no other spores in the canker, such as the brown *Sphæropsis* spores. Inoculations were immediately made with the spores which had formed in the pustules of the cankers produced on the apple limbs, using healthy apples.

Several days thereafter these apples showed all signs of the bitter rot, and after three days quantities of bitter-rot conidia were produced in the characteristic manner already described. These results proved that the fungus growing in these cankers was actually *Glomerella rufomaculans*.[a] Some of the more striking cases of artificial cankers are reproduced on Plate IX. Figures 2, 3, and 4 show artificial cankers, with control cuts above. Figure 5 shows one of the cankers (fig. 4) somewhat enlarged, with numerous black pustules.

These results showed beyond question that the bitter-rot fungus *actually produces the cankers on apple limbs*, and is not merely present in cankers which are produced by some other fungus. There was no question as to the purity of the cultures used for inoculating the apple limbs, and as there was no growth in any of the control cuts there is no reason for doubting that the fungus inoculated into the cuts on apple limbs grew in the bark and formed the pustules containing spores, which in turn produced the bitter rot in apples.

A further proof that the fungus produced the cankers was brought forward by the discovery (by Spaulding, December 24, 1902[b]) of perithecia and asci, with ascospores (Pl. V, fig. 7) of *Glomerella rufomaculans*, in one of the cankers artificially produced on apple limbs by inoculating conidia of *Glœosporium fructigenum* Berk. into apple limbs. Spaulding found that many of the spores in the artificial cankers were curved very much like the ascospores of *Glomerella rufomaculans* obtained in cultures on apple agar. The conidia and ascospores of this fungus look very much alike—so much so that it is not easy to separate them with any degree of accuracy. The only difference which is at all evident is that the conidia are usually straight, while the ascospores are slightly curved. (Pl. V, fig. 6.) Sections of the canker made by Spaulding showed the presence of perithecia (Pl. V, figs. 5 and 7), most of which were empty. Several were found, however, with asci and ascospores. (Pl. V, fig. 7.) It seems that the asci in the cankers have as little permanence as do those found in cultures. Clinton refers to the comparatively evanescent character of the asci which he found in the cultures, and we can testify to the same statement. So far as could be determined, the perithecia and asci in the cankers were identical with those of *Glomerella rufomaculans* formed in cultures. Up to this writing no perithecia or asci have been reported from the cankers on apple limbs in the orchard.

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[a] A preliminary note announcing the proof of a causal relation between the bitter-rot fungus (*Glœosporium fructigenum* Berk.) and the apple cankers was published in Science, **16**: 699, 1902.

[b] See Science, **17**: 188, 1903.

Summing up the evidence now at hand as to the causal relationship between the bitter-rot fungus (*Glomerella rufomaculans*) and the apple cankers found in certain apple orchards in Illinois and other States, we find (1) that conidial spores of this fungus which will produce the bitter rot in apples occur with great regularity in the cankers; (2) that such conidial spores taken either directly from diseased apples or from pure cultures made from cankers or diseased apples, when inoculated into the living bark of growing apple-tree branches, will produce apple cankers resembling those found in the orchards; (3) that conidial spores and asci and ascospores are contained in such artificially produced cankers, which, when inoculated into apples, produce the bitter rot.

Taken together, these facts seem to prove beyond question that the bitter-rot fungus can grow in apple branches, that by so doing it forms cankers, and that after a time spores are formed in such cankers, which produce the bitter rot in apples.

Although this fact seems established at present, the writers are by no means convinced that sufficient proof is yet at hand which would warrant the statement that *all* of the cankers found in the Illinois orchards were formed by the bitter-rot fungus. The resemblance between the canker formed by the black-rot fungus *Sphæropsis malorum* Peck on apple limbs in New York (Paddock, 1899 and 1900) and the cankers formed in Illinois suggested at first that the latter were formed by the black-rot fungus. This supposition was strengthened by the almost constant presence of spores of *Sphæropsis malorum* Peck in the Illinois cankers. The black-rot fungus is known to form cankers on apple limbs with great readiness, and the black rot is a common enemy of the apple in Illinois and adjacent States. The writers are therefore of the opinion that it is not at all improbable that the Illinois cankers are formed in part by *Sphæropsis malorum* Peck. The bitter-rot fungus may get into the young Sphæropsis cankers and both fungi may grow side by side. Considerable additional work will have to be done with these cankers before their exact identity will have been established.

SPREAD OF THE BITTER ROT.

Nearly all those who have studied the bitter rot agree that the fungus is very erratic in its time of appearance. The disease may be prevalent year after year in a particular part of an orchard without occurring in other places in the same orchard or in other orchards in the immediate locality. It has been repeatedly noted that the rot seemed to start in one tree or a group of trees close together and that it spread from the center to adjoining trees.

Observation showed that the rot was more likely to appear on trees which had once had the disease than on trees previously free from it.

One apple grower carefully marked the trees which were affected with bitter rot in 1900. When the rot first appeared in his orchard in 1902 (there was hardly any rot in 1901) he went over the orchard and found that every tree marked in 1900 had bitter rot, and not only that, but that the rot was at first confined to these trees.

Another observation frequently recorded is that the disease often starts on a few trees and, starting from this center, it gradually spreads year after year and finally affects the entire orchard. Mr. J. W. Beach, of Batavia, Ark., stated a typical case:

> I came to this country in 1884, and that season there were four trees in my old orchard affected. * * * For the last three years the disease has steadily increased, so that this year (1887) my old orchard of seventy-five trees will not yield 25 bushels of sound apples.

This apparently erratic behavior of the bitter rot can be explained in part since the discovery of the canker stage of the fungus. After its introduction into an orchard or on one tree the fungus attacks one or more branches, probably early in the summer, and produces a canker. The next year the spores from this canker will be washed down on the ripening fruit by a rain. The water is sprayed from the branch on which the canker is situated to the lower branches in the form of a cone, and one or more spores will probably fall on every apple within such a cone.

The presence of the winter stage of the fungus will explain why the rot is apt to recur on the trees affected the year before with the bitter rot, and also why the disease should first appear on such trees. The cankers produce spores early in the season, and from the trees which have cankers the disease spreads to neighboring trees.

The bitter rot is apt to appear in virulent form only once in two or three years. During the intervening periods there may be little or no rot in any one region. This may possibly be caused by weather conditions generally unfavorable to the fungus, as was the case in the summer of 1901, but it may also be due to conditions unfavorable to the growth of the fungus in the cankers.

The exact conditions which favor the development of the bitter-rot fungus on the branches are not known as yet, but it is conceivable that these might be such as would retard its growth in the canker to such an extent that few or no spores would be found during one year.

That the spores of the bitter-rot fungus are spread to the fruit from the cankers in the tree now seems proved beyond doubt. The dissemination of spores from the cankers probably begins early in summer and continues until the apples are fully grown.

Another source of infection is found in the dried mummies hanging on the trees and lying on the ground under the trees. The diseased apples of one season either fall to the ground (which most of them do) or they remain on the trees, where they dry and shrivel up. When

examined in the spring many of these mummified fruits are found to contain spores of the bitter-rot fungus in quantity. Inoculations made by us with such spores have shown them to be fully capable of remaining alive over winter and of producing the disease in July and August of the following year.

It was formerly supposed that the fungus passed the winter in the mummies, but as most of these were on the ground it was difficult to understand how the apples high up in the trees became infected. It now seems probable that the mummies play a comparatively small part in serving as distributing points for spores from year to year.

After the fungus becomes started on a number of fruits it spreads to neighboring apples and trees with great rapidity. The sticky nature of the spore masses precludes any theory of wind distribution. The spores are never dustlike, so that they could be blown about, but are generally stuck together, forming a sticky, paste-like mass. Rain and dew play an important part in distributing the spores.

Numerous small insects which frequent apple trees in the late summer, such as members of the genus Drosophila, probably carry the bitter-rot spores from one tree to another. Clinton (1902) proved that this was actually the case by placing some of these flies on sound apples and thereby producing the disease.

REMEDIAL MEASURES.

Although the bitter rot has been so destructive to apple crops for thirty years or more, little if any headway had been made until recently toward combating it successfully. The disease may still be regarded as one of the most difficult ones to control, although it now seems probable that greater success ought to attend preventive measures. These may now be placed under these heads:

1. Removal of diseased fruits and mummies.
2. Removal of limb cankers.
3. Spraying with fungicides.

REMOVAL OF DISEASED FRUITS AND MUMMIES.

Apples affected with bitter rot generally fall from the trees during the later stages of the disease. In many cases the spores in these rotted apples will live through the winter, and may be carried to sound fruits the following season. Such apples as do not fall dry up and hang on the trees all winter in a mummified condition. The next year the spores formed in these mummies may infect sound fruits. All diseased fruits on the ground should be carefully collected as soon as they fall; they should be removed from the orchard and destroyed either by drying and subsequent burning or by burying them in a deep trench, which is carefully covered with soil afterwards. Under

no circumstances should the rotted fruits be allowed to remain lying on the ground under the trees through the winter.

Dried apples on the trees should be picked and burned as soon after the fall of the leaves as possible.

Where the bitter rot appears in an orchard at isolated points it will oftentimes pay to watch the trees where the trouble first shows itself and to pick every fruit showing the slightest sign of disease. In that way the chance of having the disease spread to adjoining trees will be materially lessened.

FIG. 7.—How to cut off a large limb.

REMOVAL OF LIMB CANKERS.

It seems well established now that one of the principal sources of infection of the ripening apples is to be found in the cankers on apple limbs. These cankers should accordingly be removed and burned wherever they are found, no matter where the affected limb may be. It is often a difficult matter to find these cankers on large trees, and a good deal of patience is necessary to locate them. In cutting out the cankers the whole limb should be sawed off some distance below the cankers. Where the branch is a large one the diseased portion may be cut out without cutting off the entire limb. The best time for cutting out the cankers is during the late fall and during the winter. The cankers can be located most readily when there are no leaves on the trees. In cutting off limbs which have cankers on them, the same rules which hold for pruning branches should be observed. Small branches may be cut off with a saw at one cut. Two cuts should be made for larger branches, the first one on the under side, the second on the upper side, so as to prevent tearing off large areas of bark. (See figs. 7 and 8.)

FIG. 8.—Method of cutting a large limb which should be avoided.

All cut surfaces should be carefully trimmed, and after that they should be coated with some antiseptic substance, such as white lead paint or ordinary coal tar. The coal tar should be applied with a brush, and if too thick, it may be warmed. It is believed that by carefully removing and destroying all cankers the damage from bitter-rot infection will be very materially lessened.

SPRAYING WITH FUNGICIDES.

Prior to 1887, at a time when the bitter rot had already a wide distribution, no attempts seem to have been made to control the disease. Galloway said (1887): "With our present limited knowledge of its habits, it is impossible to suggest means for combating it." Two years later the same writer suggested spraying the fruit trees with sulphuret of potassium when the apples were about half grown, and repeating the operation every two or three weeks throughout the summer. In a brief note during the same year (1889) Galloway referred to some experiments made by Beach in 1888 with sulphuret of potassium, in which he stated that "Mr. Beach has full confidence in the remedy." Galloway (1889) reported some tests made with potassium sulphide and ammoniacal copper carbonate by Curtiss. A good many apples were saved by the spraying, little difference being noted in the results accomplished by the use of the two solutions mentioned.

Chester (1890) sprayed with potassium sulphide, liver of sulphur, and ammonium copper carbonate without decided success.

Garman (1894) published an account of a series of experiments to control diseases of apples with Bordeaux mixture. In his general summary he states:

> The proportion of rotting to nonrotting apples was in every case lessened by spraying, and we are in a position to say, as a result of these experiments, that spraying with Bordeaux mixture will save from rotting from 7¾ per cent to 31⅛ per cent of the whole number of apples. * * * The spraying increased the yield of usable apples from a little less than twofold to nearly sevenfold.

From the beginning of the nineties to the present time a number of experiments in spraying have been made by several persons, notably Alwood (1892, 1894), Stinson (1892, 1894, 1896, 1901, and 1902), Whitten (1895), Clinton (1902), Staubenrauch (1902), Burrill and Blair (1902, a and b).

Without going into details, it may be said that the results reached were favorable in some cases, but not so successful in others. Alwood (1894) states:

> Our results with Bordeaux mixture and ammonia copper carbonate, as recommended in bulletins mentioned above, are very satisfactory.

Whitten (1895) says:

> Bitter rot began first on unsprayed trees and developed more extensively than on sprayed trees. Bitter rot was less on trees sprayed with the 6-pound solution (of Bordeaux mixture) than on those sprayed with the weaker solutions.

Stinson (1896) says:

> There was much less bitter rot on the sprayed fruit than on the unsprayed. From the results obtained it was concluded that the injury can be partly prevented with three sprayings with Bordeaux mixture.

From a careful study of the results obtained up to 1900, it was evident that, although it had been shown that the disease could be

checked by spraying, there was much conflicting evidence at hand and a great many unanswered questions. Most of the experiments made were conducted for one year only. Many involved only a few trees, and many others were conducted under unfavorable conditions.

The bitter rot is a disease which has always appeared with varying virulence from year to year, sometimes destroying whole crops, then again attacking only a few apples. It has likewise made its appearance in adjoining orchards at different times in the same year. It is therefore almost impossible to draw any definite conclusions from experiments extending over one year only. Spraying in early summer may in some years check the disease should it appear only a few weeks after the spraying, while spraying at exactly the same time the next year, when the disease appears late in September, may have absolutely no effect. In order to be of value for a disease dependent upon so many varying factors as is bitter rot all spraying experiments must be conducted under exactly the same conditions in the same localities for a series of years without interruptions. Isolated tests may be and doubtless are of value in indicating what may be expected, but they are at best simply single instances, the results of which can by no means be taken as conclusive. Realizing that such was the case, a comprehensive plan for conducting spraying operations was drawn up by Mr. Waite of the Bureau of Plant Industry, U. S. Department of Agriculture, in the winter of 1900–1901, and this plan was carried out for two years in Virginia, Illinois, and Missouri, one year in cooperation with the Illinois Agricultural Experiment Station.[a] This plan was stated as follows: The object of this experiment is to answer the following questions: (1) Can Bordeaux mixture by proper spraying be made to protect apples from bitter rot? (2) Is winter treatment of the dormant trees of any assistance in the process? (3) Is early spraying more advantageous than late spraying in the treatment of this disease? (4) Are any other fungicides superior to Bordeaux mixture in the treatment of this special malady? To answer these questions the following plan was followed. The parts of the orchards experimented in were laid off in five plats, which were sprayed as follows:

Plat 1. Winter spraying only.—Spray plat and duplicate with Bordeaux mixture before the buds swell, applying the spray until the trunk and buds are blue. In duplicates, with controls.

Plat 2. Combined winter and spring spraying.—First treatment, winter spraying before buds push; second treatment, when cluster buds are open and flower buds exposed; third treatment, when the last of the petals are falling; fourth treatment, seven to ten days later. In duplicate, with controls.

[a] This plan was described in Circular No. 43 of the Illinois Agricultural Experiment Station.

Plat 3. Early spraying without the winter treatment.—Same as above, with first treatment left off; that is, second, third, fourth, and fifth sprayings.

Plat 4. Early spraying continued until summer.—Second, third, fourth, and fifth sprayings, with four or five more treatments at intervals of two weeks until about the middle of August.

Plat 5. Late spraying.—Begin when the fruit is about an inch in diameter and spray four or five times. These sprayings may be on the same dates as the last four or five treatments of plat 4.

Before giving the results obtained from the spraying in Missouri and Illinois, it may be well to state that the bitter rot appeared in a comparatively light form during both years in which the experiments were made. The experiments illustrate once more the necessity of carrying on tests of this character for a long period of years under the same or similar conditions as previously stated. In view of the fact that the attack of the disease for the past two years was a light one in some of the orchards experimented in, the conclusions reached must be considered as preliminary ones which simply go to swell the number of isolated results of spraying operations already referred to.

Replying to the questions which are enumerated above, the following general answers may be given at this time:

(1) *Can Bordeaux mixture, by proper spraying, be made to protect apples from bitter rot?*—To a certain extent, varying from 10 to 75 per cent, Bordeaux mixture surely does prevent the ravages of the bitter rot. The extent to which it will do so will depend largely on the following factors:

(*a*) *Making the Bordeaux mixture.*—Although the literature dealing with the making of this fungicide is voluminous, there is still much of it made in an improper manner. The standard Bordeaux mixture, i. e., 6 pounds of copper sulphate, 4 pounds of lime, and 50 gallons of water, is still preferable. The copper sulphate should be dissolved in one vat; the lime in another. Only freshly slaked lime should be used, and the slaking should be brought about with small quantities of water added from time to time, so as to get an even slaking. The solutions of copper sulphate and of lime should be diluted separately and then both should be run simultaneously into a third tank. A very convenient arrangement which can be constructed anywhere is shown in figure 9. The two smaller tanks are elevated on a rough platform, high enough above the third tank to allow the solution to be turned readily into the latter. This third tank in turn is elevated sufficiently to allow the finished Bordeaux mixture to run into the tanks on the spraying wagon without pumping. Such an arrangement can be built at the edge of the pond or well, from which the water for the spraying mixture is obtained. A force pump drives the necessary water into the upper smaller tanks. Great care should be

taken in weighing the proper amount of chemicals. The custom of guessing at the amounts invariably results in a poor mixture. Stock solutions of the copper sulphate, 1 or 2 pounds to the gallon, can be made and kept without deterioration, evaporation being considered.

(*b*) *On the thoroughness with which spraying is done.*—Only nozzles which throw a fine spray should be used. The Vermorel is the standard nozzle for good work. In spraying, the old precept that all parts of a tree should be reached by the spray holds good. With a fungus like that of the bitter rot, where a large number of spores may reach a fruit, thorough spraying alone will be effective. The apparent failure to control the bitter rot in many instances is doubtless due to the fact that even with the best spraying it is impossible to cover the

FIG. 9.—Arrangment of vats used in making Bordeaux mixture.

entire fruit with the fungicide. Numerous fruits were found on sprayed trees where the fungus had started to grow between points on the fruit covered with the fungicide. It is very desirable that further information concerning the number of sprayed fruits actually rotted be obtained.

(2) *Is winter treatment of the dormant trees of any assistance in the process?*—The spraying of trees before the buds opened showed no apparent benefit. This, however, ought not to be taken as conclusive, especially in view of the recent discovery of a stage in the life history of the bitter-rot fungus in the wood and bark of the apple tree. In the absence of any data as to the exact time when this bark stage—the canker—is reached, it may be that winter spraying will to some extent check the development of possible cankers.

(3) *Is early spraying more advantageous than late spraying in the treatment of this disease?*—No evidence was obtained from the experiments calculated to prove that early spraying is more advantageous than late spraying. The later sprayings—that is, those made after the apples were half formed—in our estimation were the most effective. The early spraying resulted in a severe rusting of the fruit during one year. The tissues of the young fruits developed very slowly under the spots where the fungicide covered the epidermis, and rusted and misshapen fruits resulted in many cases. Thick-skinned varieties like the Ben Davis showed this rusting to a marked extent. The exact explanation for this rusting as a result of spraying with Bordeaux mixture is still to be determined.

(4) *Are any other fungicides superior to Bordeaux mixture in the treatment of this special malady?* The Bordeaux mixture is, as far as our tests go, certainly superior to any of the other fungicides used, i. e., ammonium copper carbonate and potassium sulphide.

Summing up the evidence from our own spraying operations as well as those of others, the following recommendations can be made at this time:

(1) For spraying to prevent bitter rot use the standard Bordeaux mixture, i. e., 6 pounds of copper sulphate, 4 pounds of lime, and 50 gallons of water.

(2) Spray at least once before the buds open. This spraying of the trunks and branches should be thorough, so as to reach any possible spores about to form cankers or coming from cankers.

(3) Spray during the middle of the summer, beginning about the middle of June, and continue several times until the fruit is almost ripe. Should the attack come very late in the summer, use the ammonium copper carbonate, so as to avoid damaging the fruit for the market, because of a possible lime deposit from the Bordeaux mixture.

In addition to these sprayings aimed especially at bitter rot, it is necessary to spray with Bordeaux mixture to which Paris green or some arsenical poison has been added just after the petals have fallen and again ten days to two weeks later. This is directed against apple scab, codling moth, and leaf-eating insects, as well as bitter rot and leaf blight.

SUMMARY AND RECOMMENDATIONS.

The facts presented in the foregoing pages may briefly be summarized as follows:

(1) The bitter rot or ripe rot is one of the most serious diseases of apples. The loss due to this disease in 1900 was estimated (for the United States) as $10,000,000. It is one of the most difficult diseases to control and is constantly on the increase.

(2) The bitter rot is due to a fungus, *Glomerella rufomaculans* (Berk.)

Spaulding & von Schrenk, hitherto generally known as *Glœosporium fructigenum* Berk.

(3) This fungus until 1902 was known only in its conidial stage on pomaceous fruits and grapes. The perfect or ascus stage has since been discovered both in cultures on fruits and in artificial cankers on the apple limbs.

(4) The fungus attacks ripening apples during July and August, and is most virulent during moist, hot summers. It is most active on apples in the belt of States on the line of the Ohio River, from Virginia on the Atlantic Ocean to Oklahoma in the West, and southward. (See fig. 1.)

(5) During the past summer canker-like areas were discovered on apple limbs from which the disease seemed to spread. These cankers generally occurred in the upper parts of trees and contained spores of the bitter-rot fungus, as proved by direct inoculations into apples.

(6) Inoculations into healthy apple limbs of bitter-rot spores from pure cultures of the bitter-rot fungus (made both from diseased apples and cankers) resulted in the formation of cankers similar to those found in the orchards. Spores from these cankers produced the bitter rot in sound fruits. This proves beyond doubt that the bitter-rot fungus is the cause of the cankers on apple limbs in the orchard.

(7) The spores of the bitter-rot fungus are washed from the cankers onto the apples below the cankers. Spores are carried from tree to tree by insects, and possibly by raindrops.

(8) One of the best methods for combating this disease will consist in carefully cutting out all cankers during the winter. These should be burned at once. All diseased apples on the ground or in the tree should be collected and destroyed. As a further precaution, trees should be sprayed with standard Bordeaux mixture at least once before the buds open, and again frequently from midsummer until the fruits are almost ripe.

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DESCRIPTION OF PLATES.

PLATE I. Frontispiece. Apples affected with bitter rot resulting from inoculation from a canker. Two rings of spore pustules have already formed in the apple shown at the top of the plate.

PLATE II. Typical examples of apples affected with bitter rot. These fruits were inoculated with spores from a diseased apple.

PLATE III. A view in a Missouri apple orchard during a severe epidemic of bitter rot. Almost the entire crop of apples is diseased and has fallen from the trees within a few days.

PLATE IV. Figs. 1, 3, and 7.—Apples attacked by the bitter-rot fungus at various stages of the disease. Fig. 7 shows an apple with a rotten spot only 2 or 3 days old; fig. 3, probably four or five days after the attack; and fig. 1, a little later. They all show how the diseased area becomes sunken, and later on wrinkled. They also show the rings of spore masses. Fig. 2.—An apple affected with bitter rot. The small dark spots show the appearance of early stages of this disease. Fig. 4.—A mummified apple which was attacked by the bitter-rot fungus in summer, and remained on the tree the following winter. It is dried up and shriveled. The spore masses dried, but are still present in quantities in the mummy, as shown in the figure. This illustrates the necessity for removing the mummies. Fig. 5.—Two pears, of which the one on the left was inoculated at one point with bitter-rot spores and the other pear not inoculated. After several weeks the inoculated fruit (which remained on the tree) showed a typical bitter-rot spot, with well-developed spores of the bitter-rot fungus. Fig. 6.—A small piece of a winter squash inoculated with spores of the bitter-rot fungus. The fungus grew readily on the squash, and after a brief period numerous pustules with spores formed, as shown in the figure.

PLATE V. Fig. 1.—A mass of spores of the bitter-rot fungus growing in a pure culture. These spores form in countless thousands in such a culture for a period of 8 to 14 days. Fig. 2.—Various stages of germinating bitter-rot spores. Some have but one germ tube, others have two. On some of the threads dark secondary spores form. Fig. 3.—Starch grains in various stages of solution by the ferment given off by the fungus. The fungus threads growing in the cells of the apple digest the starch grains. Fig. 4.—Peculiar spore-like bodies formed by germinating spores of a cactus fungus (*Glœosporium cactorum*), which resemble the black bodies shown in fig. 2. Figs. 5 and 7.—The perfect or perithecial stage of the bitter-rot fungus as found during the winter of 1902–3 in the apple cankers. Fig. 5 shows two empty perithecia from an apple canker; fig. 7 a single perithecium much enlarged with some of the asci in place. Fig. 6.—A group of three mature asci with ascospores of the bitter-rot fungus and one immature ascus. These asci were developed in a pure culture of the bitter-rot fungus. The ascospores so much resemble the ordinary spores that it is difficult to distinguish them.

PLATE VI. Fig. 1.—A pure culture on apple agar, showing the luxuriant development of the bitter-rot fungus, with large numbers of spores. After a time this spore production stops and peculiar hard black masses develop. These masses are shown in Fig. 1. From a number of them large drops of a yellowish liquid are exuding. Each black mass contains one or more perithecia of the bitter-rot fungus, with asci, such as are shown on Plate V, fig. 6. Fig. 2.—Enlarged group of pustules on a diseased apple. On quiet, moist nights the spore masses of the bitter-rot fungus exude from the mouths of the pustules on the apple fruit in long filmy threads. Some of these threads, composed wholly of spores, are shown exuding from some of the pustules. Figs. 3 and 4.—Apple diseased with bitter rot and control fruit. These figures show the result of an experiment to determine whether the spores found in the apple cankers would produce the bitter rot in apples. A pure culture was made with spores taken from an apple canker. Spores from this pure culture were inoculated into an apple (fig. 3), while another apple was punched full of holes with a sterilized needle. As shown in the figures, the inoculated fruit (fig. 3) developed a typical case of the bitter rot, while the control fruit (fig. 4) was still sound. This is one of numerous proofs that spores from the apple cankers produce the disease.

PLATE VII. Three typical cankers such as are now believed to be formed on apple limbs during one stage in the life cycle of the bitter-rot fungus. Spores from these cankers produce the bitter rot of apples. Such cankers should be cut out and burned wherever found.

PLATE VIII. Cankers on living apple limbs. Fig. 1.—A number of cross sections of apple cankers, illustrating the manner in which the wood turns brown immediately under the dead bark. It also shows the healing callus forming at the edge of the dead areas. The largest section shows two such healing layers, proving that this canker is at least two years old. Figs. 2 and 4.—Apple branches, fig. 4 showing probably early stages of canker. Fig. 3.—A branch swollen at the point where an apple was borne the previous year. Many such were found to contain bitter-rot spores. It is possible that the fruit spurs became infected from diseased fruits through the stems of the apples.

PLATE IX. Young apple cankers produced by inoculating spores from pure cultures of the bitter-rot fungus into bark slits on healthy apple trees. In all cases here shown small cankers were formed, pustules with spores developed, and the spores, when inoculated into apples, produced the bitter rot of the fruit. This formed the last link in the chain of evidence necessary to show the connection between the bitter-rot fungus and the apple canker.

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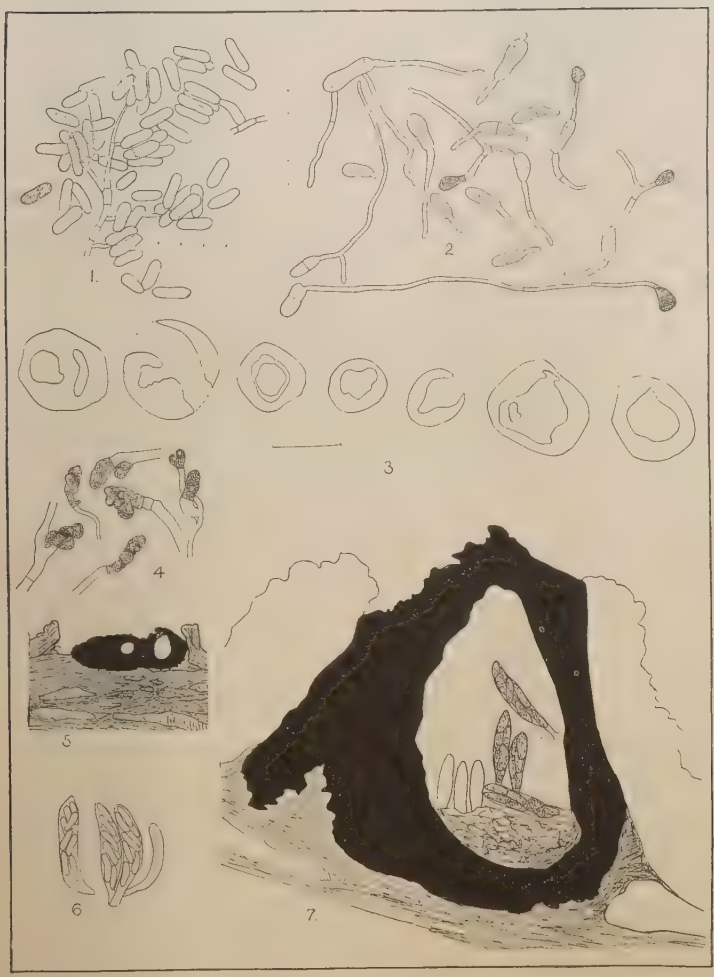
APPLES AFFECTED WITH BITTER ROT.

Inoculation from a diseased apple.

DISEASED APPLES UNDER TREES.

THE BITTER-ROT FUNGUS ON VARIOUS FRUITS.

1, 3, and 7. Various stages of growth on apples; 2, An early stage of the disease; 4, A mummified apple from the preceding year; 5, Growth of the bitter rot fungus on pear, with control; 6, Growth of the bitter-rot fungus on squash.



VARIOUS STAGES OF THE BITTER-ROT FUNGUS.

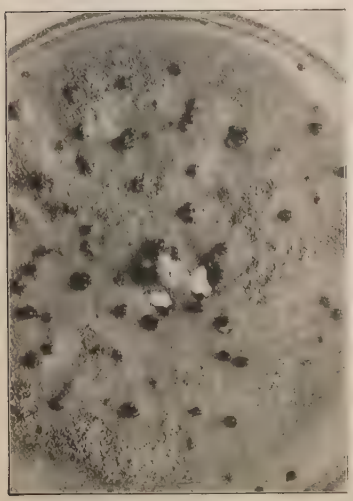


FIG. 1.—PLATE CULTURE.

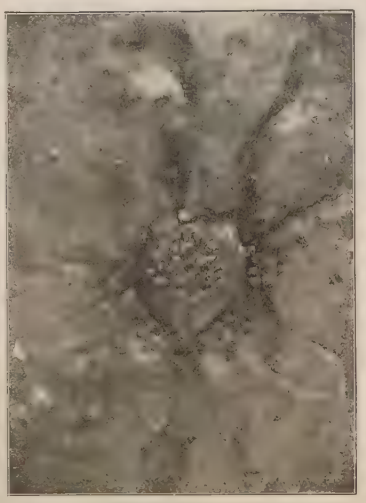


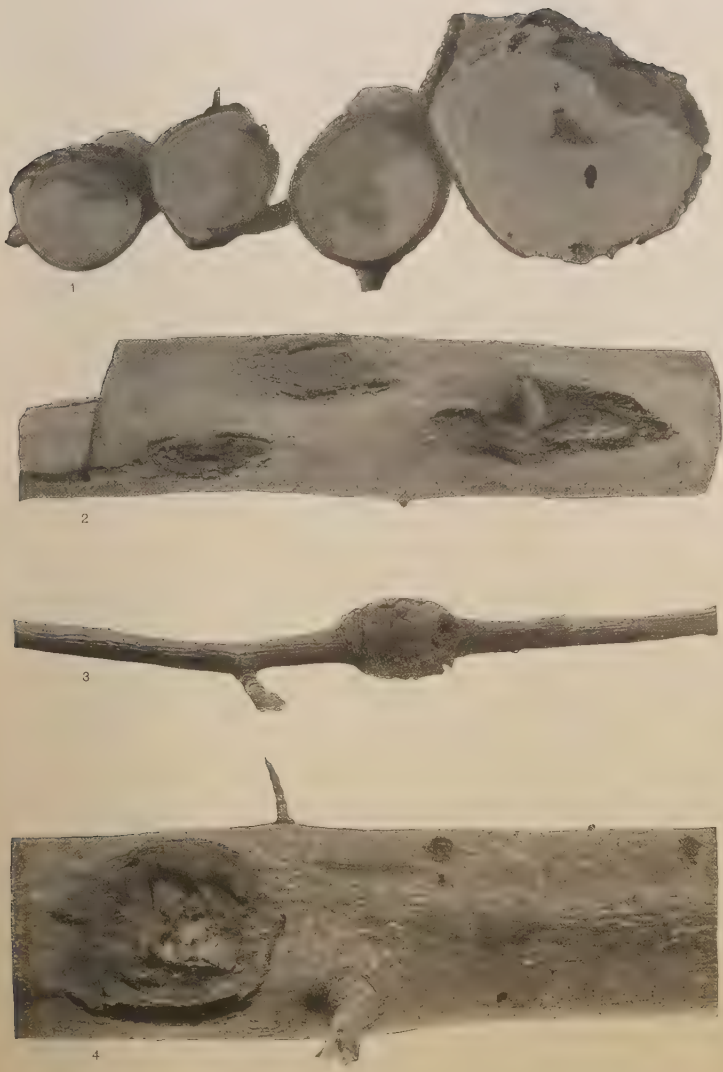
FIG. 2.—PUSTULES ON APPLE.

FIG. 3.—APPLE INOCULATED FROM A PURE CULTURE OBTAINED FROM A CANKER.

FIG. 4.—CONTROL FRUIT

STAGES OF GROWTH OF THE BITTER-ROT FUNGUS.

THREE LIMBS WITH BITTER-ROT CANKERS FROM LIVING APPLE TREES.



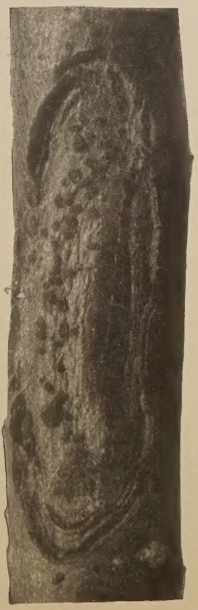
BITTER-ROT CANKERS ON LIVING APPLE LIMBS.

1

2

3

4



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ARTIFICIAL CANKERS PRODUCED ON LIVING APPLE LIMBS BY INOCULATION WITH SPORES OF THE BITTER-ROT FUNGUS.

[Continued from p. 2 of cover.]

No. 22. Injurious Effects of Premature Pollination; with General Notes on Artificial Pollination and the Setting of Fruit without Pollination. 1902. Price, 10 cents.
23. Berseem: The Great Forage and Soiling Crop of the Nile Valley. 1902. Price, 15 cents.
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26. Spanish Almonds and Their Introduction into America. 1902. Price, 15 cents.
27. Letters on Agriculture in the West Indies, Spain, and the Orient. [In press.]
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